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Genetic evaluation for bovine tuberculosis resistance in dairy cattle

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INTERPRETIVE SUMMARY

Bovine Tuberculosis (bTB) is a chronic disease of grave consequences to the dairy and beef cattle sector. A genetic evaluation platform was developed to estimate the genetic merit of animals with regards to bTB resistance. The presence of significant genetic variation rendered the distinction between genetically susceptible and resistant animals possible. Genetic evaluations for bTB resistance are now official in Great Britain.

ABSTRACT

Genetic evaluations for resistance to bovine tuberculosis (bTB) were calculated based on British national data including individual animal tuberculin skin test results, post-mortem examination (presence of bTB lesions and bacteriological culture for *Mycobacterium bovis*), animal movement and location information, production history and pedigree records. Holstein cows with identified sires in herds with bTB breakdowns (new herd incidents) occurring between the years 2000 and 2014 were considered. In the first instance, cows with a positive reaction to the skin test and a positive post-mortem examination were defined as infected. Values of zero and one were assigned to healthy and infected animal records, respectively. Data was analyzed with mixed models. Linear and logit function heritability estimates were 0.092 and 0.172, respectively. In subsequent analyses, breakdowns were split into two-month intervals to better model time of exposure and infection in the contemporary group. Intervals with at least one infected individual were retained and multiple intervals within the same breakdown were included. Healthy animal records were assigned values of zero, and infected records a value of one in the interval of infection and values reflecting a diminishing probability of infection in the preceding intervals. Heritability and repeatability estimates were 0.115 and 0.699, respectively.

Reliabilities and across time stability of the genetic evaluation were improved with the interval model. Subsequently, two more definitions of "infected" were analyzed with the interval model: (i) all positive skin test reactors regardless of post-mortem examination; (ii) all positive skin test reactors plus non-reactors with positive post-mortem examination. Estimated heritability was 0.085 and 0.089, respectively; corresponding repeatability estimates were 0.701 and 0.697. Genetic evaluation reliabilities and across time stability did not change. Correlations of genetic evaluations for bTB with other traits in the current breeding goal were mostly not different from zero. Correlation with the UK Profitable Lifetime Index was moderate, significant and favorable. Results demonstrated the feasibility of a national genetic evaluation for bTB resistance. Selection for enhanced resistance will have a positive effect on profitability and no antagonistic effects on current breeding goal traits. Official genetic evaluations are now based on the interval model and the last bTB trait definition.

Key words: Genetic evaluation, bovine tuberculosis resistance

INTRODUCTION

Bovine tuberculosis (bTB) is a chronic bacterial disease of cattle caused *by Mycobacterium bovis* (*M. bovis*) infection primarily involving the respiratory tract. The disease affects animal health and welfare, causing substantial financial strain to the dairy cattle sector worldwide through involuntary culling, animal movement restrictions and the cost of control and eradication programs (Allen et al, 2010). Furthermore, bTB is considered a zoonotic disease with considerable public health implications in countries where it is not subject to mandatory eradication programs.

In Great Britain, the majority of bTB cases are recorded in south western England and Wales. A bTB control and eradication program has been in place in these areas since 1950 comprising primarily routine and targeted surveillance of cattle herds, culling of positive animals and movement restrictions on infected herds. Surveillance is based on the administration of the single intradermal comparative cervical tuberculin test (skin test) involving two separate injections of sterile purified mixtures of *M. avium* and *M. bovis* antigens (tuberculins) in the deep layer of the skin of the neck, followed by examination of the skin for localized allergic reactions after 72 hours (de la Rua-Domenech et al, 2006). When reaction to the *M. bovis* tuberculin injection is deemed to be less than or equal to that to the *M. avium* tuberculin injection, then the skin test is considered negative (non-reactor). A positive skin test result (known as a reactor) is declared when the reaction to *M. bovis* tuberculin exceeds that to *M. avium* tuberculin by more than 4 mm, according to the standard international interpretation (de la Rua-Domenech et al, 2006). In all other cases the test is considered inconclusive and repeated 60 days later. If one or more animals in a herd react positively to the skin test then a new bTB incident, also known as breakdown, is declared prompting animal movement restrictions, suspension of the official bTB free (OTF) status of the herd, and systematic testing of all animals in the herd at 60-day intervals. Animals with a positive or two consecutive inconclusive skin tests are compulsorily slaughtered and examined at the abattoir for visible lesions of bTB in their organs. Tissue samples from a representative number of infected animals from each herd are submitted to the laboratory to isolate *M. bovis* in bacteriological culture. A positive post-mortem examination result (presence of lesions and/or positive *M. bovis* culture*)* signals a downgrading of the herd's OTF status from "suspended" to "withdrawn". The breakdown remains open and skin testing continues in the herd

until one or two (depending on the post-mortem results and location of the herd) consecutive negative tests at minimum intervals of 60 days are obtained on all remaining animals.

Implementation of bTB control and eradication programs incurs significant costs to taxpayers on an annual basis. During 2010-2011, these costs amounted to £152 million in Great Britain and £23 million in Northern Ireland (Abernethy et al, 2013). However, despite the investment and good control efforts, the incidence and prevalence of bTB cases in Great Britain constantly increased between the mid-1980s and 2012, although they have leveled-off in more recent years. Even so, just over 4,800 new breakdowns were declared in cattle herds and more than 36,000 animals had to be slaughtered for bTB control purposes in 2015 (Department for Environment Food and Rural Affairs - DEFRA, 2016). This has been partly attributed to a reservoir of endemic *M. bovis* infection in wildlife, especially badgers, in large parts of England and Wales. All these facts hinder progress towards achieving the DEFRA's goal for Great Britain to be OTF by year 2038.

The presence of genetic variation among individual animals in their immunological response to *M. bovis* exposure was documented by Pollock et al (2002). This genetic variation was subsequently quantified and moderate heritability estimates were reported in cattle (Bermingham et al, 2009; Brotherstone et al, 2010; Tsairidou et al, 2014). The amount of genetic variation and the level of estimated heritability render resistance to bTB amenable to improvement via genetic selection. Breeding for enhanced bTB resistance could complement existing control and eradication programs. However, relevant tools have not been widely available as no formal genetic evaluation systems have been put in place.

The objective of the present study was to assess the feasibility of a national genetic evaluation for bTB resistance in dairy cattle based on British population data. We combined data from

various sources and developed automated data handling procedures suitable for a routine commercial process. We investigated different models and trait definitions.

MATERIALS AND METHODS

Data

Population surveillance data were made available from the Animal and Plant Health Agency (APHA) of the Department for Environment, Food and Rural Affairs (DEFRA). Data consisted of tuberculin skin test and post-mortem examination records of dairy and beef cattle from Great Britain (predominantly England and Wales), spanning the period 1957-2014 although more than 90% of the recorded data were post 2000. Skin tests had been applied to individual animals every two months within a given breakdown (defined as the period of disease surveillance in a herd prompted by the first detection of an infected animal and ending with the lifting of herd movement restrictions). Animals were classified as non-reactors, inconclusive reactors and reactors as described by de la Rua-Domenech et al (2006).

Negative skin test results for individual animals (non-reactors) were not being systematically recorded in the APHA database prior to 2011. Therefore, the British Cattle Movement Service (BCMS) database was used to identify contemporaries of reactors and inconclusive reactors in the APHA database that were present in the same herd during each breakdown. All contemporaries found in the BCMS database that were not included in the APHA data were considered to be non-reactors. The combined APHA-BCMS data was merged with milk recording data to derive information about the date of calving and parity number of the animals. A final match with the national pedigree dataset (including data from the official Herdbooks) maintained by the Edinburgh Genetic Evaluation Services on behalf of the Agriculture and

Horticulture Development Board (Dairy), retrieved the identification of the sire of each cow. Figure 1 illustrates the combination of data from various sources. A total of 5,358,308 cow records were included in the initial project database.

Trait Definition

The health status of each animal was defined as follows:

1. Infected; three definitions were examined:
    a. Reactors to the skin test with positive post-mortem examination results comprising visible lesions of bTB and/or positive *M. bovis* culture (R+PM); this conservative definition required that a positive skin test be confirmed post-mortem and is consistent with the current formal APHA definition of a confirmed case as well as a previous study based on similar data (Brotherstone et al, 2010).
    b. All reactors to the skin test regardless of post-mortem examination results (R); this definition was based on the very high specificity (ca. 99%) and positive predicted value of the skin test (de la Rua-Domenech et al, 2006; Goodchild et al, 2016) implying a very small percentage of false positives (positive skin test reactors that were not actually diseased).
    c. As in (b) plus non-reactors and inconclusive reactors to the skin test who had been subsequently slaughtered and had positive post-mortem examination results (RandNPM); this definition aimed at capturing all information available that could be indicative of infection including possible false negative skin test reactors in the analysis (Allen et al, 2010).
2. Healthy: live non-reactors to the skin test or slaughtered non-reactors with negative post-mortem examination results (i.e. absence of lesions and a negative *M. bovis* culture).

Based on the above, three trait definitions of the animal's bTB infection status were considered according to the three definitions of "infected". The "healthy" animal definition was the same in all cases.

Data Edits

More than 90% of the records in the database were from breakdowns that started in the year 2000 or later. The latter data were also more complete in terms of post-mortem examination results. Therefore, breakdowns that started before 2000 were removed from further analyses. This edit was consistent with a previous study conducted on similar data (Brotherstone et al, 2010). Additional edits kept only milking cows of the Holstein breed with an identified Holstein sire in breakdowns that were not shorter than two months. A final edit required that breakdowns have at least five observations of which at least one pertained to an infected cow. According to the three trait definitions, data from 424,843; 642,995 and 660,762 daughters of 15,211, 19,050 and 19,325 sires, respectively, were kept in the analysis.

Genetic Evaluation

In the first instance, the following animal model was used to analyze animal bTB infection status as defined above:

$$Y_{ijkmn} = \mu + B_i + R_j \cdot M_k + L_m + b_1 dur + b_2 age + b_3 phol + A_n + e_{ijkmn} \quad (1)$$

where

Y = bTB infection status record of animal n in breakdown i (0/1)

μ = population mean

B = fixed effect of the breakdown i

R·M = fixed effect of the interaction between calendar year j and month k of breakdown onset

L = fixed effect of lactation number m (m=1 for primiparous cows, 2 for multiparous cows)

dur = linear regression on duration of the breakdown (b_1=regression coefficient)

age = linear regression on age of animal at breakdown onset (b_2=regression coefficient)

phol = linear regression on percentage of Holstein genes of the animal (b_3=regression coefficient)

A = random additive genetic effect of animal n including pedigree (6,398,839 animals)

e = random residual

Although data were restricted to only Holstein cows, the percentage of Holstein (vs. British Friesian) genes was available in the national dairy pedigree and was included in the model, consistent with the national genetic evaluations for other traits (Edinburgh Genetic Evaluation Service, 2016).

In a separate analysis, a logit function was fitted to model 1 to account for the binary nature of the trait.

In model 1, the entire breakdown irrespective of length represented a contemporary group (cohort of animals). Although the model adjusted for different breakdown duration, the time of exposure and actual infection could vary considerably within and across breakdowns, thereby affecting the true definition of the contemporary group and possibly impacting on results. In an alternative design, breakdowns were split into equally-sized (two months) intervals that would better capture the specific prevailing conditions and dynamics at a given time, and model

exposure and infection consistently within and across breakdowns and herds. The interval duration of two months was chosen in connection with bimonthly surveillance testing of herds during open breakdowns. As before, a breakdown interval was required to have at least one infected animal and a minimum size of five to be included in the analysis. Data from multiple intervals within the same breakdown were included, resulting in repeated records per individual cow. Specifically, animals defined as healthy in a given interval were assumed to have been healthy in all previous intervals within the same breakdown and were assigned repeated records of zero. An animal found to be infected in a given interval was assigned a record of one in this interval. In previous intervals within the same breakdown, this infected animal was assigned a value reflective of a diminishing probability of infection manifested as a record of $(0.40)^n$, where n was the time distance from the interval of infection; for example, the infected animal record was 0.40 in the immediately previous interval, 0.16 in the interval before that, 0.064 in the third preceding interval and so on. The probability of infection chosen (0.40) is consistent with a sensitivity estimate of 0.60 of the skin test as diagnostic tool for bTB. Sensitivity reflects the proportion of negative skin test reactors (non-reactors) that were truly healthy; thus the value of 0.40 represents the proportion of diseased non-reactors (false negatives). Reported sensitivity estimates of the tuberculin skin test range in literature from 0.51 to 0.81 (Downs et al, 2011; Álvarez et al, 2012; Karolemeas et al, 2012). Varying the assumed sensitivity and probability of infection between these values had only trivial impact on the genetic evaluation results (data not shown).

The model of analysis under the interval design was revised as follows:

$$Y_{ijklmno} = \mu + B_i + R_j \cdot M_k + L_m + D_l + b_1 age + b_2 phol + A_n + PE_n + e_{ijklmno} \qquad (2)$$

where

Y = bTB infection status record of animal n in breakdown interval i (repeated records)

B = fixed effect of the breakdown interval i

R·M = fixed effect of the interaction between calendar year j and month k of breakdown interval onset

D = fixed effect of breakdown interval duration l (l=1 for a two-month interval, 2 for a possibly shorter interval leading to the end of the breakdown)

age = linear regression on age of animal at breakdown interval onset (b_1=regression coefficient)

PE = random permanent environment effect associated with animal n

All other effects were as in model (1).

In all cases, variance component and parameter estimates were derived using the software ASReml (Gilmour et al, 2009) and genetic evaluations (estimation of breeding values) with the software MiX99 (Vuori et al, 2006). Reliability estimates of the genetic evaluations, reflecting the squared correlation between the estimated and true breeding values, were based on the approximation proposed by Jamrozik et al (2000). Variance component estimation was based on a subset of data pertaining to sires with 20 to 500 daughters in the data. This edit resulted in about one third of the data being used in variance component estimation, in each case.

Separate genetic evaluations were calculated after removing the last two years of data and repeating the analyses on the reduced dataset. Results from the reduced and full data analyses were compared to test the stability of the genetic evaluation across time by emulating conditions of consecutive genetic evaluations with updated data. Additional model validation was conducted based on Interbull's method 3 for national genetic evaluations, which entails

regression of current (full) on the previous (reduced) genetic evaluation and on a function of the number of new daughters per sire since the previous evaluation (Boichard et al, 1995). This function combines the number of new daughters by year of first calving with the total number of daughters in the current evaluation (Boichard et al, 1995).

RESULTS AND DISCUSSION

Descriptive Statistics

Table 1 summarizes the three datasets considered in the present study, depending on trait definition. In the breakdown design (model 1) each cow had a single record whereas repeated records were included in the interval design (model 2). It should be noted that these proportions reflect only breakdowns with infected cases included in the present study and are not representative of the entire national herd.

As expected, the conservative definition of infection (R+PM, requiring a positive post-mortem examination of skin test reactors) resulted in the lowest proportion of infected animals (3.57%). There was minimal difference between the other two datasets which were mainly based on all skin test reactors regardless of post-mortem results (8.28% vs. 8.29%). The last dataset also included non-reactors and inconclusive reactors that had been slaughtered and tested positively post-mortem. However, there were very few such cases; in fact, of all infected cases in the third dataset (RandNPM), 97.3% were skin test reactors, 2.6% were inconclusive and only 0.1% were non-reactors to the skin test.

Breakdown vs. Interval Model

Results from the breakdown design (model 1) and the interval design (model 2) were compared using the first trait definition (R+PM), where skin test reactors with positive post-mortem were

considered to be infected. The heritability estimates were 0.093 (±0.009) and 0.115 (±0.014) for the two models, respectively. Heritability estimate after fitting a logit function to model 1 was 0.172 (±0.018), reflecting the genetic variation in the underlying liability scale. These estimates are in agreement with results of previous studies on British (Brotherstone et al., 2010) and Irish (Bermingham et al., 2009) bTB data considering the same trait definition. Presence of significant ($P<0.01$) genetic variance signifies the amenability of the trait to improvement via selective breeding. Model 2 also yielded a repeatability estimate of 0.699 (±0.005) indicative of the definition of repeated records of the same cow within a breakdown in the present study.

Figure 2 shows the histogram of sire estimated breeding values (EBVs) by models 1 and 2. In accordance to industry preference, positive numbers were associated with higher resistance to bTB. Both models yielded normally distributed sire EBVs. The average proportion of infected daughters among the top and bottom 20 bulls from the evaluation based on the breakdown model was 2% and 23%, respectively. Corresponding proportions for the interval model were 2% and 24%, respectively. Thus the two models fared equally well at distinguishing sires whose offspring have a higher degree of resistance from those that are more susceptible.

Table 2 summarizes the reliability estimates of sire EBVs obtained by the two models. Results are expressed as the cumulative percentage of sires falling within each reliability range. For example, 78% and 90% of the sires had EBV reliability greater than or equal to 0.30 based on the breakdown and interval model, respectively. Proportionally, more than twice the number of sires had EBV reliability of at least 0.50 based on the interval compared to the breakdown model, whereas this proportion was trebled for higher reliabilities (≥0.60). The average sire EBV reliability was 0.40 and 0.54 for the breakdown and interval model, respectively. These results

attest to the increased accuracy on the interval model, reflecting a more appropriate definition of the contemporary group and a larger amount of data in the genetic evaluation.

Figure 3 illustrates the relationship between sire EBVs and proportion of infected daughters in the genetic evaluation. In both models, sire EBVs were reflective of the infection rate among their daughters, with somewhat stronger correlations for the interval than the breakdown model (-0.68 vs. -0.64). These correlations are expectedly negative as a higher EBV is indicative of increased resistance to bTB manifested by a lower infection rate.

Stability of genetic evaluations across time is illustrated in Figure 4. In both cases, sire EBVs based on a reduced data set were very good predictors of EBVs based on full data, the latter emulating a future genetic evaluation including new records. In this research case, new records were from an additional two full years of bTB surveillance, adding more than 30% of new data to the genetic evaluation. Official national genetic evaluations in the UK are calculated three times per year meaning new data will be included more gradually leading to even higher correlations and stability between successive evaluation runs. High EBV correlations and stability across time are crucial for the acceptability of genetic evaluation results by the industry.

Validation with Interbull method 3 yielded a significantly greater than zero ($P<0.01$) regression on the function of new daughters for the breakdown model but a non-significant one ($P=0.29$) for the interval model. If a genetic evaluation is unbiased, this regression is expected to be zero (Boichard et al, 1995). Furthermore, Interbull require the regression to not exceed 0.02 genetic standard deviations in order to include a national genetic evaluation in their international comparisons (www.interbull.org). In the present study, the regression in question was 0.0338 and 0.0053 genetic standard deviations for the breakdown and the interval model, respectively, making the latter acceptable for national genetic evaluations.

The above results collectively demonstrate an overall superiority of the interval over the breakdown model in the analysis of bTB data. Therefore, further analyses were based on the former.

Comparison of Trait Definitions

The interval model was used to analyze data based on the other two trait definitions, where all skin test reactors (R) and all skin test reactors plus non-reactors with positive post-mortem (RandNPM), respectively, were considered to be infected.

Table 3 summarizes the variance component and heritability estimates from the three interval model analyses. All estimates were statistically greater than zero ($P<0.01$). Slightly higher heritability was estimated for the conservative definition of infected (R+PM), which can be attributed to the lower estimates for residual and permanent environmental variance (Table 3). The latter may be due to the definition of the trait, which, combined with the requirement to include breakdown intervals with at least one infected record, resulted in fewer records per cow compared to the more relaxed definitions (R and RandNPM). In fact, the average number of records per cow increased from 2.45 in R+PM to 3.38 and 3.47 for the other two definitions, respectively (Table 1). In all cases, genetic variance was of equal size and significant ($P<0.01$) attesting to the amenability of all traits to genetic improvement via selection.

The distribution of sire EBV based on the R and RandNPM trait definitions was similar to those in Figure 2 for the interval model (R+PM). Table 4 illustrates differences between the top 20 and bottom 20 sires, by EBV, in the three genetic evaluations. Sires with a minimum EBV reliability of 0.30 and daughters in at least 10 breakdowns were considered in this Table. The distinction between the best and worst sires was more pronounced in the R and RandNPM cases compared to the conservative definition (R+PM). This can be attributed to the more relaxed definition in

331 the last two cases, allowing more infected individuals to be included in the analysis. Enhanced
332 capacity to distinguish sires by their genetic merit is expected to facilitate genetic progress.

333 Average reliability of sire EBV was 0.54, 0.54 and 0.55 for the three trait definitions (R+PM, R
334 and RandNPM), respectively. The distribution of sires across ranges of EBV reliability was very
335 similar to the interval model results shown in Table 2 for the conservative definition (R+PM).
336 The advantage of the larger amount of data and increased progeny group size in the last two
337 definitions (33.8 and 34.2 daughters per sire, respectively) compared to R+PM (27.9) was
338 seemingly offset by the increased heritability of the latter (Table 3).

339 Product moment correlations between sire EBVs based on the three trait definitions are shown in
340 Table 5. As expected, correlations were strongest between the last two definitions considering all
341 skin test reactors (R and RandNPM). Weaker correlations with R+PM can be primarily attributed
342 to the number of diseased animals that reacted positively to the skin test and were culled without
343 having had the time to develop and exhibit post-mortem lesions.

344 The stability of genetic evaluations across time was tested for all trait definitions and results
345 were very similar to those in Figure 2. Correlations between reduced and full model EBV were
346 0.94, 0.95 and 0.95 for R+PM, R and RandNPM, respectively. Validation with the Interbull
347 method 3 yielded very similar results in R and RandNPM analyses to those for R+PM described
348 above. In all cases, the genetic evaluations were shown to be unbiased as far as this method is
349 concerned.

350 Correlations between sire EBV for bTB with the interval model and official EBV for other traits
351 in the current national breeding goal are shown in Table 6. Sire EBV with a minimum reliability
352 of 0.30 and daughters in minimum 10 herds (2,039-2,996 sires, depending on trait definition)

were considered for this purpose. These results illustrate the generally weak and favorable correlation between genetic evaluations for bTB and other important traits. The strongest correlation estimates (0.15) was with the overall Profitable Lifetime Index (£PLI), which effectively combines all economically important traits in one single value (Agriculture and Horticulture Development Board, 2016). Significant ($P<0.05$) correlations were also observed with lifespan, which describes the functional longevity of a cow, reflecting the probability of being involuntarily culled after adjusting for milk yield. Relatively stronger correlations pertaining to R and RandNPM can be attributed to losses of animals that react positively to the skin test and have to be culled, regardless of the outcomes of post-mortem examination. These estimates indicate that selection for increased resistance to bTB may have small favorable effects on £PLI and cow longevity. In general, Table 6 suggests that no antagonistic effects on animal traits already in the breeding program should be expected from sire selection for enhanced bTB resistance. This is consistent with the UK £PLI placing over 65% of its emphasis on health traits.

The availability of bTB resistance genetic evaluations provides the industry with a number of options to add to the existing control measures. Farmers may choose to avoid particularly poor bulls when another bull of similar £PLI is available. Breeding companies may make only desirable bulls available in high risk areas and may incorporate bTB in their bull dam choices where possible. These choices combined and made over time would be expected to lead to a general reduction in the infection rate in UK herds.

The bTB evaluations are now being used to create genomic breeding values. At the cow level, genomic breeding values would allow farmers to exclude young animals at an early age if they were predicted to be particularly susceptible to bTB. For example, if farmers removed the worst 5% of their animals each year before they had a chance to infect the remainder of the herd, the

expectation would be that the overall level of herd infectivity would decrease over time and, therefore, the potential of each animal to infect another would be reduced. Similarly, the potential of a herd to pass infection to wild reservoirs would be reduced, thereby further decreasing the overall level of infectivity in the population. The genetic epidemiology of such a proposed policy warrants further study to determine an optimal strategy for the use of genetic evaluations in reducing overall bTB infection.

CONCLUSIONS

The feasibility of a genetic evaluation for enhanced bTB resistance using nationally available data was demonstrated in the present study. Results have shown that selective breeding can potentially make a positive contribution (when used alongside other interventions such as cattle movement restrictions and biosecurity improvements) to DEFRA's stated aim for Great Britain to be OTF by 2038.

As of January 2016, the interval model has been applied in the official national genetic evaluation of Holstein sires considering all reactors to the skin test plus non-reactors and inconclusive reactors with positive post-mortem results as infected individuals. Further work is planned to address bTB resistance in the other dairy breeds as well as beef cattle.

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REFERENCES

Abernethy D.A., P. Upton, I. M. Higgins, G. McGrath, A. V. Goodchild, S. J. Rolfe, J. M. Broughan, S. H. Downs, R. Clifton-Hadley, F. D. Menzies, R. de la Rua-Domenech, M. J. Blissitt, A. Duignan and S. J. More. 2013. Bovine tuberculosis trends in the UK and the Republic of Ireland, 1995–2010. *Veterinary Record* 172:312 doi:10.1136/vr.100969.

Agriculture and Horticulture Development Board, UK. 2016. http://dairy.ahdb.org.uk/technical-information/breeding-genetics/%C2%A3pli/#.Vx-XH3p8qRQ.

Allen A.R., G. Minozzi, E.J. Glass, R.A. Skuce, S.W.J. McDowell, J.A. Woolliams and S.C. Bishop. 2010. Bovine tuberculosis: the genetic basis of host susceptibility. *Proc. Royal Society B-Biological Sciences* 277, 2737-2745. PubMed PMID: PMC2981996.

Álvarez J., A. Perez, J. Bezos, S. Marqués, A. Graue, J.L. Saez, O. Mínguez, L. de Juan and L. Domínguez. 2012. Evaluation of the sensitivity and specificity of bovine tuberculosis diagnostic tests in naturally infected cattle herds using a Bayesian approach. *Veterinary Microbiology* 155 (1): 38–43.

Bermingham M.L., S.J. More, M. Good, A.R. Cromie, I.M. Higgins, S. Brotherstone and D.P. Berry. 2009. Genetics of tuberculosis in Irish Holstein-Friesian dairy herds. *J Dairy Science* 92:3447–3456.

Boichard D., B. Bonaiti, A. Barbat and S. Mattalia. 1995. Three methods to validate the estimation of genetic trend for dairy cattle. *J Dairy Science* 78:431-437.

Brotherstone S., I.M.S. White, M. Coffey, S.H. Downs, A.P. Mitchell, R.S. Clifton-Hadley, S.J. More, M. Good and J.A. Woolliams. 2010. Evidence of genetic resistance of cattle to infection with *Mycobacterium bovis*. *J Dairy Science* 93: 1234-1242.

Department for Environment, Food and Rural Affairs, UK. 2016. https://www.gov.uk/government/uploads/system/uploads/attachment_data/file/529314/bovine tb-statsnotice-quarterly-15jun16.pdf.

Downs S.H., J. Parry, J. Nunez-Garcia, D.A. Abernethy, J.M. Broughan, A.R. Cameron, A.J. Cook, R. de la Rua-Domenech, A.V. Goodchild, M. Greiner, J. Gunn, S.J. More, S. Rhodes, S. Rolfe, M. Sharp, H.M. Upton, H.M. Vordermeier, E. Watson, W. Welsh and A.O. Whelan. 2011. Meta-analysis of diagnostic test performance and modelling of testing strategies for control of bovine tuberculosis. *Proc. Society for Veterinary Epidemiology and Preventive Medicine* 139-153.

Edinburgh Genetic Evaluation Service. 2016. http://www.sruc.ac.uk/info/120275/egenes.

Gilmour A.R., B.J. Gogel, B.R. Cullis and R. Thompson. 2009. ASReml User Guide Release 3.0 VSN International Ltd, Hemel Hempstead, HP1 1ES, UK www.vsni.co.uk.

Goodchild A.V., S.H. Downs, P. Upton, J.L.N. Wood and R. de la Rua-Domenech. 2016. Specificity of the comparative skin test for bovine tuberculosis in Great Britain. *Veterinary Record* doi:10.1136/vr.102961.

Jamrozik J., L.R. Schaeffer and G.B. Jansen. 2000. Approximate accuracies of prediction from random regression models. *Livest. Prod.* Sci. 66: 85–92.

Karolemeas K.R. R. de la Rua-Domenech, R. Cooper, A.V. Goodchild, R.S. Clifton-Hadley, A.J.K. Conlan, A.P. Mitchell, R.G. Hewinson, C.A. Donnelly, J.L.N. Wood and T.J.

McKinley. 2012. Estimation of the relative sensitivity of the comparative tuberculin skin test in tuberculous cattle herds subjected to depopulation. *PLoS ONE* 7(8): e43217. doi:10.1371/journal.pone.0043217.

Pollock J. M. and S.D. Neill. 2002. *Mycobacterium bovis* infection and tuberculosis in cattle. *Veterinary Journal* 163 (2): 115-127.

de la Rua-Domenech R., A.V. Goodchild, H.M. Vordermeier, R.G Hewinson, K.H. Christiansen and R.S. Clifton-Hadley. 2006. Ante mortem diagnosis of tuberculosis in cattle: A review of the tuberculin tests, gamma-interferon assay and other ancillary diagnostic techniques. *Research in Veterinary Science* 81, 190-210.

Tsairidou S., J.A. Woolliams, A.R. Allen, R.A. Skuce, S.H. McBride, D.M. Wright, M.L. Bermingham, R. Pong-Wong, O. Matika, S.W.J. McDowell, E.J. Glass and S.C. Bishop. 2014. Genomic prediction for tuberculosis resistance in dairy cattle. *PLoS ONE* 9(5): e96728. Epub 2014/05/09. PubMed PMID: 24809715; PubMed Central PMCID: PMCPmc4014548.

Vuori K., I. Strandén, M. Lidauer and E.A. Mäntysaari. 2006. MiX99 - effective solver for large and complex linear mixed models. *Proc. 8th World Congress on Genetics Applied to Livestock Production*, Belo Horizonte, Minas Gerais, Brazil, 13-18 August, 2006.

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Table 1. Three datasets in the genetic evaluation according to bTB trait definition[1].

	R+PM	R	RandNPM
No. cows	424,843	642,995	660,762
No. records*	1,040,891	2,170,322	2,294,859
No. sires of cows	15,211	19,050	19,325
No. breakdowns	4,365	8,158	8,397
No. breakdown intervals*	7,585	18,079	18,822
Prop. infected cows	0.0357	0.0828	0.0829

[1]R+PM: bTB infected = skin test reactors with positive post-mortem results; R: bTB infected = all skin test reactors regardless of post-mortem results; RandNPM: bTB infected = as R plus non-reactors and inconclusive reactors with positive post-mortem results.

*Interval model only.

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Table 2. Reliability of sire genetic evaluations[1] based on the breakdown and interval models; cumulative percentage of sires per reliability range.

Reliability range	Breakdown model	Interval model
< 0.10	100%	100%
0.10 - 0.19	94%	97%
0.20 - 0.29	89%	94%
0.30 - 0.39	78%	90%
0.40 - 0.49	42%	73%
0.50 - 0.59	22%	53%
0.60 - 0.69	12%	37%
0.70 - 0.79	7%	25%
0.80 - 0.90	4%	13%
$\geq$ 0.90	2%	6%

[1]bTB infected = skin test reactors with positive post-mortem results.

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Table 3. Variance components and parameter estimates (est.) and standard errors (s.e.)[1] from the interval model analyses.

	R+PM		R		RandNPM	
	est.	**s.e.**	**est.**	**s.e.**	**est.**	**s.e.**
Genetic variance	0.006	0.001	0.006	0.001	0.007	0.001
Permanent environment variance	0.032	0.001	0.047	0.001	0.046	0.001
Residual variance	0.016	<0.001	0.023	<0.001	0.023	<0.001
Phenotypic variance	0.055	<0.001	0.076	<0.001	0.076	<0.001
Heritability	0.115	0.014	0.085	0.007	0.089	0.007
Repeatability	0.699	0.005	0.701	0.002	0.697	0.002

[1]R+PM: bTB infected = skin test reactors with positive post-mortem results; R: bTB infected = all skin test reactors regardless of post-mortem results; RandNPM: bTB infected = as R plus non-reactors and inconclusive reactors with positive post-mortem results.

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Table 4. Differences between top 20 and bottom 20 sires in genetic evaluations based on three datasets[1] and the interval model; sires with minimum reliability of 0.30 and daughters in at least 10 herds were considered.

	R+PM	R	RandNPM
Difference in % of infected daughters	22%	33%	35%
Difference in estimated breeding values	0.17	0.21	0.21

[1]R+PM: bTB infected = skin test reactors with positive post-mortem results; R: bTB infected = all skin test reactors regardless of post-mortem results; RandNPM: bTB infected = as R plus non-reactors and inconclusive reactors with positive post-mortem results.

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Table 5. Product-moment correlations between genetic evaluations (above diagonal) and number of common bulls (below diagonal) based on three data definitions[1] and the interval model.

	R+PM	R	RandNPM
R+PM		0.62	0.64
R	14,998		>0.99
RandNPM	15,201	19,050	

[1]R+PM: bTB infected = skin test reactors with positive post-mortem results; R: bTB infected = all skin test reactors regardless of post-mortem results; RandNPM: bTB infected = as R plus non-reactors and inconclusive reactors with positive post-mortem results.

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Table 6. Genetic evaluation correlations between bovine tuberculosis[1] and other traits.

Trait	R+PM	R	RandNPM
Milk Yield	0.00	0.05	0.06
Fat Yield	-0.02	0.08*	0.08*
Protein Yield	0.01	0.10*	0.10*
Fat %	-0.02	0.02	0.01
Protein %	0.02	0.07*	0.06
Milk Somatic Cell Count	-0.04	-0.05	-0.06
Fertility Index[2]	0.03	0.05	0.05
Calving Interval	0.00	-0.03	-0.03
Conception Rate	0.06	0.06	0.05
Calving Ease (direct)	0.06	0.08*	0.08*
Calving Ease (maternal)	0.04	0.06	0.07*
Lifespan	0.07	0.10*	0.11*
Profitable Lifetime Index	0.06	0.15*	0.15*

[1]R+PM: bTB infected = skin test reactors with positive post-mortem results; R: bTB infected = all skin test reactors regardless of post-mortem results; RandNPM: bTB infected = as R plus non-reactors and inconclusive reactors with positive post-mortem results.

[2]Combination of calving interval and non-return in 56 days.

*P<0.05. Positive correlations are favorable except for Milk Somatic Cell Count and Calving Interval.

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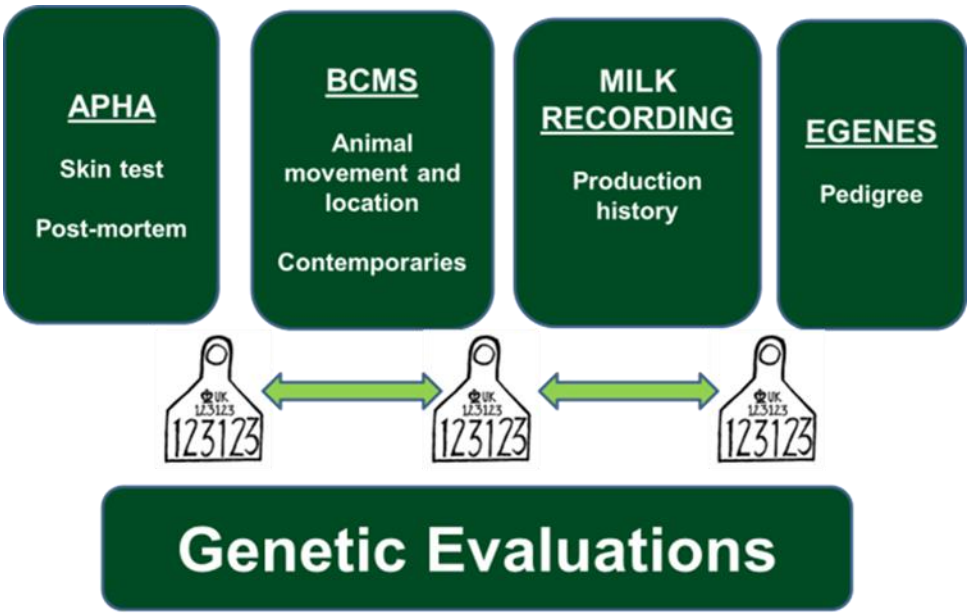
Figure 1. Combination of data from different sources in the genetic evaluation for bTB resistance; APHA=Animal and Plant Health Agency; BCMS=British Cattle Movement Service; EGENES= Edinburgh Genetic Evaluation Services.

Figure 2. Histogram of sire estimated breeding values (EBV) based on the breakdown and interval models; bTB infected = skin test reactors with positive post-mortem results.

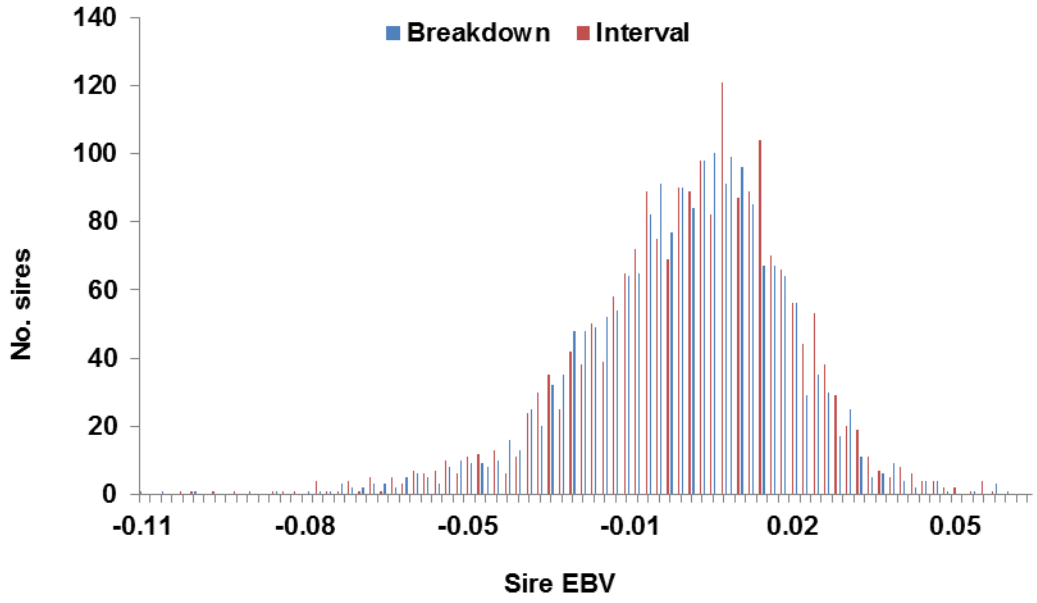
Figure 3. Sire estimated breeding values (EBVs) plotted against the proportion of infected daughters on which EBVs were based, using the breakdown and interval models; r=correlation; bTB infected = skin test reactors with positive post-mortem results.

Figure 4. Sire genetic evaluations based on the full dataset (vertical axis) plotted against genetic evaluations based on the reduced dataset (minus last two years, 30% less), using the breakdown and interval models; r=correlation between genetic evaluations; bTB infected = skin test reactors with positive post-mortem results.

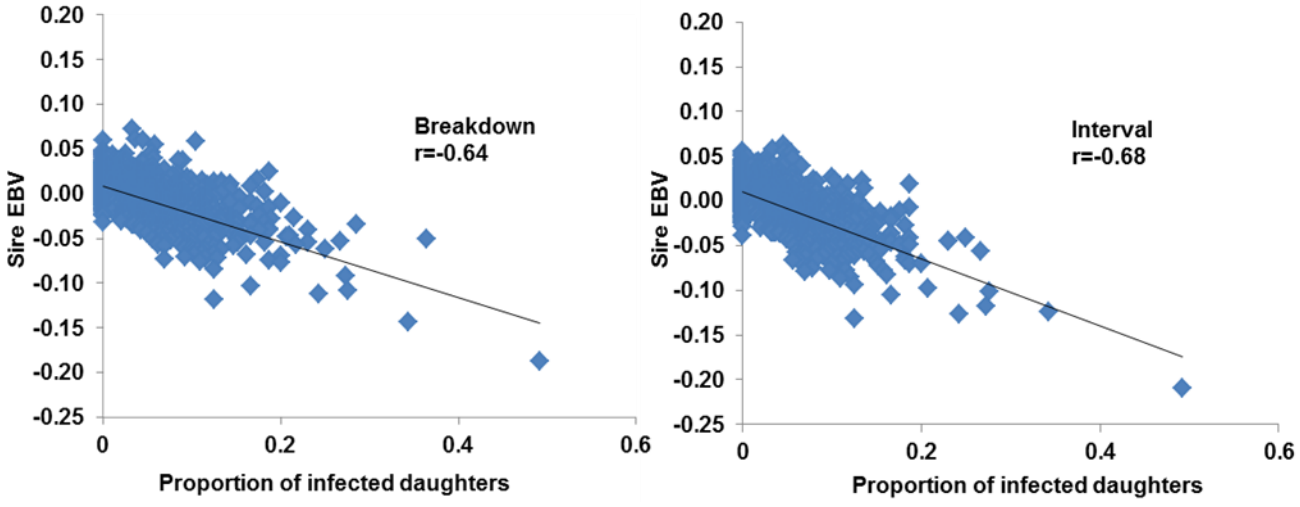
Banos Figure 1



Banos Figure 2



491 **Banos Figure 3**



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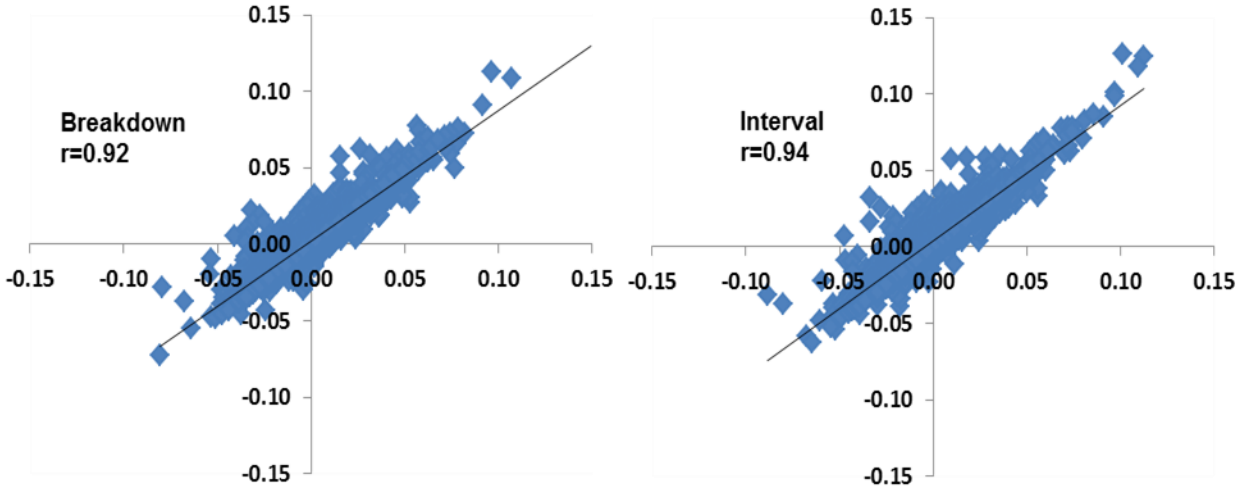
494 **Banos Figure 4**



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